

ANTI-HIV-1 DRUGS

(A JiTT Session Reading Assignment)

Learning Objectives

By the end of the lecture you should be able to:

1. Indicate the drugs (antiretrovirals) used for treatment of HIV-1 infection.
2. Explain the rationale for using HAART combinations and toxicities observed with their long-term use.
3. Define the following concepts for understanding of clinical decision making:
 - Viral replication and suppression
 - Viral drug-resistance
 - Adherence to treatment
 - Immune reconstitution
 - Complications of treatment
4. Describe the current guidelines for the treatment of HIV-infected patients.
5. Identify the circumstances that change the guidelines.

Drug List:

Reverse Transcriptase Inhibitors (RTIs):

NRTIs (Nucleoside RTIs): zidovudine, stavudine, lamivudine, didanosine, abacavir, combivir.

NtRTIs (Nucleotide RTIs): tenofovir, adefovir.

NNRTIs (Non-nucleoside RTIs): nevirapine, delavirdine, and efavirenz.

Protease Inhibitors: saquinavir, indinavir, nelfinavir, amprenavir, lopinavir, lopinavir / ritonavir, atazanavir, darunavir.

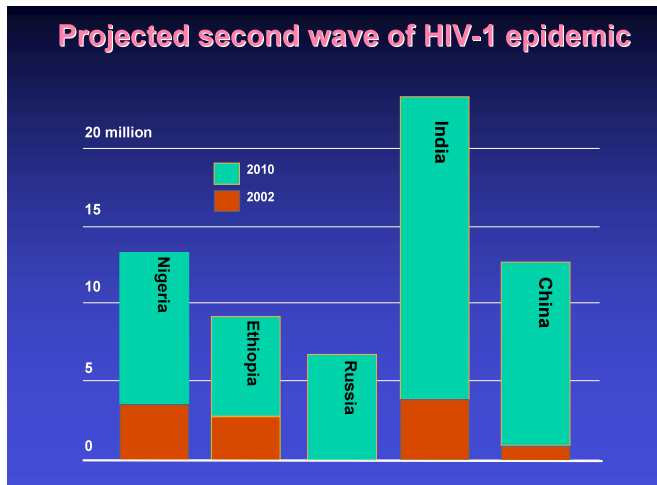
Entry Inhibitors: enfuvirtide, maraviroc.

Integrase Inhibitor: raltegravir.

Common Abbreviations:

HIV-1:	Human Immunodeficiency virus type-1
AIDS:	Acquired Immune Deficiency syndrome
RTIs:	Reverse Transcriptase Inhibitors
NRTIs:	Nucleoside Reverse Transcriptase Inhibitors
NtRTIs:	Nucleotide Reverse Transcriptase Inhibitors
NNRTIs:	Non-nucleoside Reverse Transcriptase Inhibitors
HAART:	Highly Active Antiretroviral Therapy

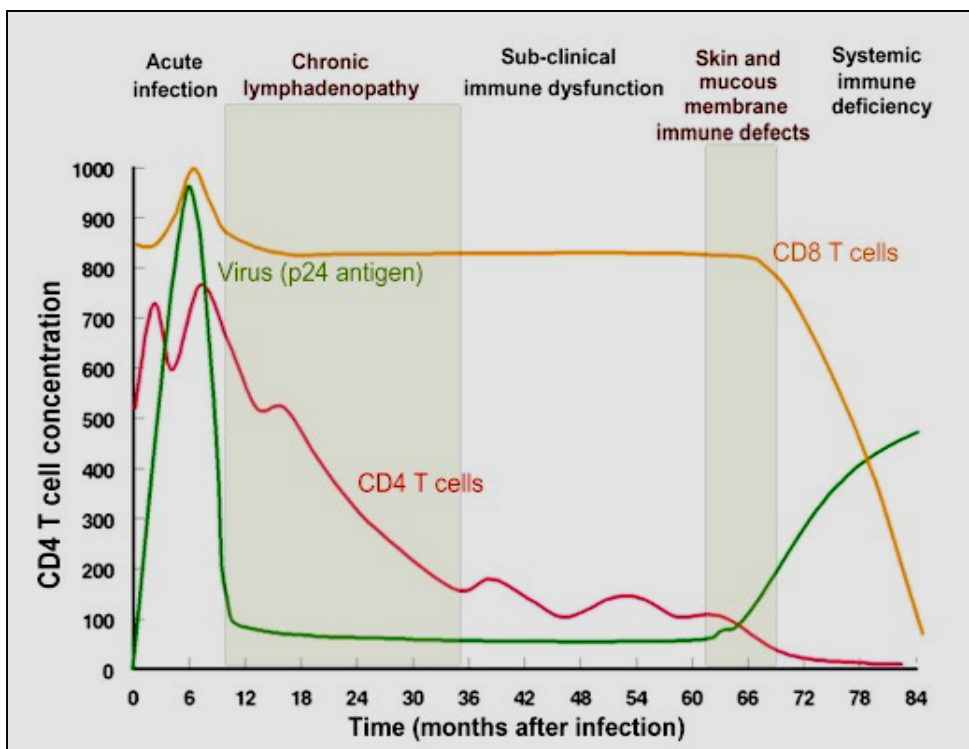
Introduction: Since it was first recognized in 1981, the death toll from Acquired Immune Deficiency Syndrome (AIDS) has surpassed 50 million worldwide. Despite recent improvements in both



availability and access to antiretroviral treatment and care in many regions of the world, the AIDS pandemic killed an estimated 2.1 million people, including 330,000 children, in 2007. The pandemic is not homogeneous within regions, with some countries more afflicted than others. Despite the implementation of prevention strategies, the number of people infected with HIV continues to rise in most parts of the world. Sub-Saharan Africa is by far the worst-affected region, with an estimated 22.5 million at the end of 2007 -- 68% of the global total. In addition, South & Southeast Asia have an estimated 12% of the global total, and a projected 2nd wave of the epidemic has already started in China and India.

Following Human immunodeficiency virus type-1 (HIV-1) infection, the immune system begins to fail, leading to life-threatening opportunistic infections and progression towards AIDS. HIV primarily infects vital cells in the immune system such as T-cells, monocytes and macrophages. HIV replication in helper T-cells (specifically CD4⁺ T-cells) is highly productive and cytopathic. When CD4⁺ T-cell numbers decline below a critical level, cell-mediated immunity is lost, and the body becomes progressively more susceptible to opportunistic infections and malignancies. Eventually most HIV-infected individuals develop AIDS. Indeed, the stage of infection can be determined by measuring the patient's CD4⁺ T-cell count, and the level of HIV in the blood. Without treatment, about 9 out of every 10 persons with HIV will progress to AIDS within 10–15 years. However, many progress much sooner.

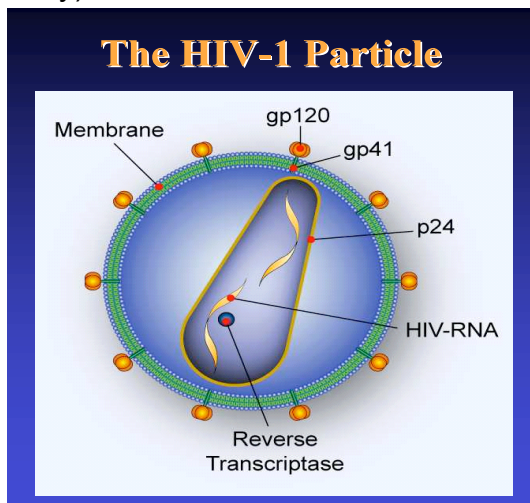
Basically, HIV infection has four stages: Early incubation period, acute infection, latency stage, and AIDS.



The initial incubation period upon infection is asymptomatic and usually lasts between two and four weeks. The second stage, acute infection, lasts an average of 28 days and can include symptoms such as fever, lymphadenopathy, pharyngitis, rash, myalgia, malaise, and esophageal sores. The latency stage, which occurs third, shows few or no symptoms and can last anywhere from two weeks to upto twenty years and beyond. AIDS, the fourth and final stage

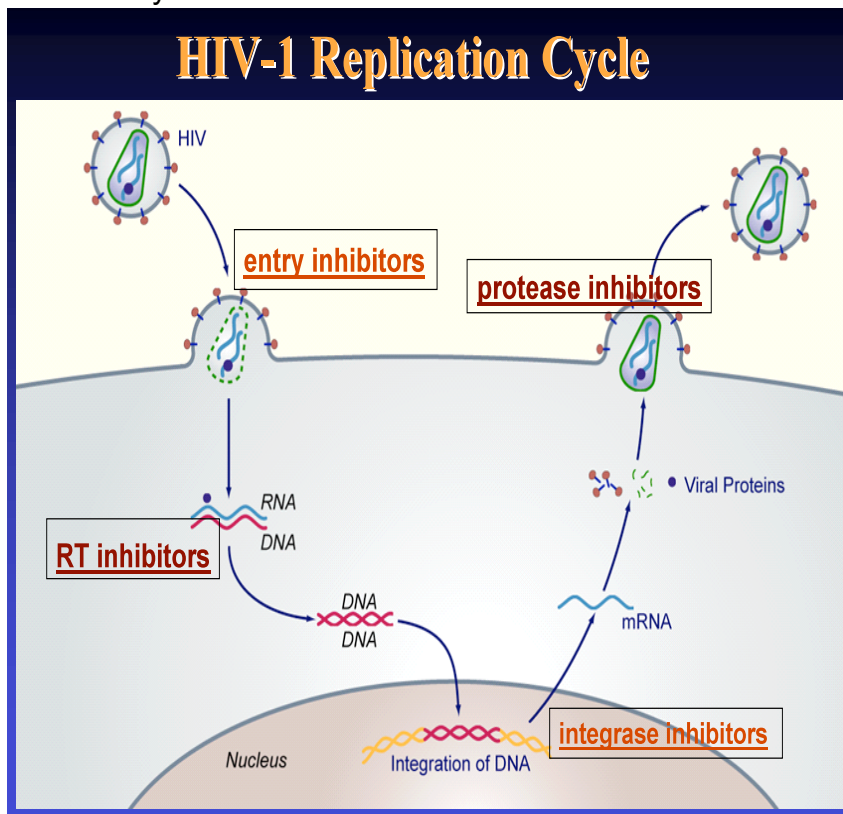
of HIV infection, shows as symptoms of various opportunistic infections associated with a progressive decrease of the CD4⁺ T cell count and an increase in viral load. Treatment with antiretrovirals increases the life expectancy of people infected with HIV. The average survival time with antiretroviral therapy (as of 2005) is estimated to be more than 5 years, and without antiretroviral therapy, death normally occurs within a year after the onset of AIDS.

Human immunodeficiency virus type-1 (HIV-1) is a lentivirus (a member of the retrovirus family) which are known to cause slow chronic diseases. **The HIV-1 viral particle** is composed of



two copies of positive single-stranded RNA that codes for the virus's nine genes enclosed by a conical capsid composed of the viral protein p24. The single-stranded RNA is tightly bound to nucleocapsid containing the virion proteins; reverse transcriptase, protease and integrase. A matrix surrounds the capsid which is in turn surrounded by the viral envelope. The envelope is composed of two layers of phospholipids (taken from the host cell membrane when a virus particle buds out). The viral envelope consists of trimeric proteins that are encoded by the Env gene. The bud is composed of glycoprotein (gp) 120, and the stem consists of three gp41 molecules. This glycoprotein complex enables the virus to attach to and fuse with target cells to initiate the infectious cycle.

Infectious cycle: HIV enters cells by the binding (adsorption) of glycoproteins present on its surface (gp160) to receptors, both CD4 and a chemokine receptor (generally either CCR5 or CXCR4). Entry begins through interaction of the trimeric envelope complex (gp160 spike) on the cell surface followed by fusion with the cell membrane and release of the HIV capsid into the cell. Several **Entry inhibitor** drugs are available to



supress viral entry into the cells, e.g. the fusion inhibitor **enfuvirtide** and the CCR5 antagonist, **maraviroc**. The HIV RNA and various viral enzymes (reverse transcriptase, integrase, ribonuclease and protease) are injected into the cell. These steps are known as adsorption and uncoating. In the cytoplasm, the viral single strand RNA genome is then reverse transcribed into double strand DNA. The process of reverse transcription is extremely error-prone, and the resulting mutations may cause drug resistance or allow the virus to evade the body's immune system. A whole range of nucleoside and non-nucleoside analogs are currently

available as **reverse transcriptase inhibitors**, e.g. the NRTIs, NtRTIs and NNRTIs. The double stranded DNA is then transported into the nucleus which is then integrated into the host chromosome, to form the provirus. This integration of viral DNA is carried out by the viral enzyme called *integrase*. The FDA has recently approved an HIV-1 **integrase inhibitor, raltegravir**. The integrated viral DNA may lie dormant (the latent stage of HIV infection) or actively produce the virus (productive infection) and kill the host T-cells. A number of cellular transcription factors, e.g. Sp1 and NF- κ B (Nuclear factor kappa B), as well as viral transactivators (e.g. Tat protein) are essential for transcriptional activation of the provirus. During viral replication, the integrated DNA (provirus) is transcribed into a multicistronic mRNA (coding for multiple proteins), which is then spliced into smaller pieces. These small pieces are exported from the nucleus into the cytoplasm, where they are translated into the early regulatory proteins, e.g. Tat (which encourages new virus production) and Rev (facilitates the transport of viral RNAs to the cytosol). The late structural proteins Gag and Env are then produced from the full-length mRNA. The final step of the viral replication cycle, assembly of new HIV-1 virions, begins at the plasma membrane of the host cell. Here, the Gag (p55) and Gag-Pol (p160) polyproteins associate with the inner surface of the plasma membrane along with the HIV genomic RNA, as the forming virion begins to bud from the host cell. The polyproteins are then cleaved by the HIV protease and the various structural components assemble to produce a mature HIV virion. This cleavage step is essential for viral maturation and can be inhibited by the HIV **protease inhibitors**.

Highly active antiretroviral therapy (HAART): Current treatment for HIV infection consists of highly active antiretroviral therapy, or HAART. This has been highly beneficial to many HIV-infected individuals since its introduction in 1996, when the protease inhibitor-based HAART initially became available. The introduction and widespread use of HAART has converted the HIV associated disease as a chronic disease in countries where these drugs are readily available, and facilitated the recovery of those patients who have access to the health care system and treatment. However, majority of the world's HIV-infected individuals do not have access to these expensive medications.

In developed countries, most of the current research has been directed towards the improvements of current treatments, includes decreasing side effects of current drugs, further simplifying drug regimens to improve adherence, and determining the best sequence of regimens to manage drug resistance.

Current HAART options are combinations (or "cocktails") consisting of at least three drugs belonging to at least two "classes" of antiretroviral agents. Typically, these classes are two nucleoside analogue reverse transcriptase inhibitors (NRTIs) plus either a protease inhibitor (PI) or a non-nucleoside reverse transcriptase inhibitor (NNRTI). New classes of drugs such as Entry Inhibitors and Integrase Inhibitors provide treatment options for patients who are infected with viruses already resistant to common therapies, although they are not widely available and not typically accessible in resource-limited settings.

The only known method of prevention is avoiding exposure to the virus. However, a course of antiretroviral treatment administered immediately after exposure, referred to as post-exposure prophylaxis, is believed to reduce the risk of infection if begun as quickly as possible. In developed countries where HAART is available, doctors assess their patients thoroughly: measuring the viral

load, how fast CD4 declines, and patient readiness. The physicians then decide when to recommend starting treatment.

The timing for starting HIV treatment is still debated. In the absence of HAART, progression from HIV infection to AIDS has been observed to occur at a median of between nine to ten years and the median survival time after developing AIDS is only 9.2 months. One study suggests that the average life expectancy of an HIV infected individual is 32 years from the time of infection if treatment is started when the CD4 count is 350/ μ L. However, there is no question that treatment should be started before the patient's CD4 count falls below 250, and most national guidelines say to start treatment once the CD4 count falls below 350. In addition, because AIDS progression in children is more rapid and less predictable than in adults, particularly in young infants, more aggressive treatment is recommended for children than adults.

Classification of currently available antiretroviral drugs:

Reverse Transcriptase Inhibitors (RTIs)		Protease inhibitors (PIs)	Entry inhibitors
Nucleoside analogue (NRTIs)	Non-Nucleoside analogues (NNRTIs)		
Zidovudine (ZDV)	Nevirapine	Saquinavir	Enfuvirtide
Didanosine (ddI)	Delavirdine	Ritonavir	Maraviroc
Stavudine (d4T)	Efavirenz	Indinavir	
Abacavir		Nelfinavir	
Lamivudine (3TC)		Amprenavir	
Emtricitabine		Lopinavir/ritonavir (<i>Kaletra</i>)	Integrase inhibitor
		Atazanavir	Raltegravir
Tenofovir (a NtRTI)		Fosamprenavir	

Remember, HAART neither cures the patient nor does it uniformly remove all symptoms. Indeed, rebound of high titre of HIV, often HAART resistant, return if treatment is stopped. Moreover, it would take more than a lifetime for HIV infection to be cleared using HAART. Despite this, many HIV-infected individuals on HAART have experienced remarkable improvements in their general health and quality of life, which has led to a large reduction in HIV-associated morbidity and mortality.

However, the HAART regimen most often achieves less than optimal results, due to a variety of reasons, such as medication intolerance/side effects, prior ineffective antiretroviral therapy and infection with a drug-resistant strain of HIV. Indeed, non-adherence to antiretroviral therapy is the major reason that most individuals fail to benefit from HAART. The reasons for non-adherence and non-persistence with HAART are varied. These include, complexity of the HAART regimens, such as pill number, dosing frequency, meal restrictions, etc. Other issues such as side effects of HAART, may also lead to non-adherence. The long-term side effects of HAART include lipodystrophy, dyslipidemia, insulin resistance, an increase in cardiovascular risks and birth defects.

In the following sections, discussion of the mechanism of action and the side effects of each group of drug is emphasized, followed by a thorough analysis of the complications observed with various HAART combinations.

A. Reverse Transcriptase Inhibitors (RTI):

These drugs inhibit the activity of reverse transcriptase (RT) enzyme; hence, no double stranded viral DNA is produced to be incorporated into the host cellular genome. There are two major classes of RTIs:

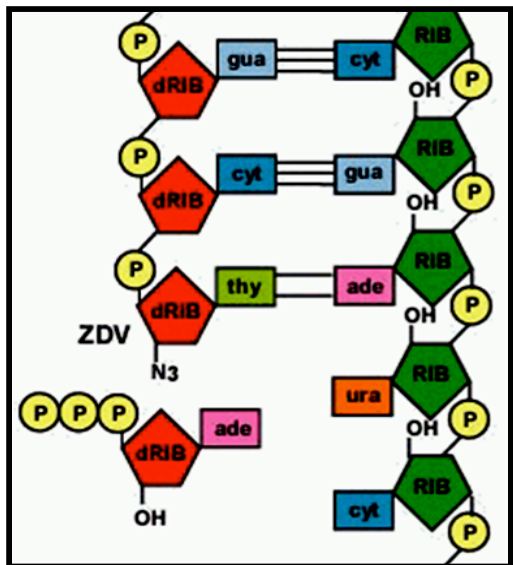
- Nucleoside/nucleotide analogues (NRTI/ NtRTI)
- Non-nucleoside analogues (NNRTI)

1. Nucleoside analogue reverse transcriptase inhibitors (NRTIs): The mechanism of action of NRTIs is competitive inhibition of the RT enzyme. They compete with the natural enzyme substrates

(deoxynucleoside triphosphates; dNTP) for binding with the RT. The NRTIs incorporate into the growing DNA chain leading to premature chain termination. They are active against both HIV-1 and HIV-2. Long intracellular half-lives of NRTIs allow for once or twice/day dosing (even for drugs not initially designed for once/day based on serum half-life). They are excreted through the kidneys and thus require dose adjustment in patients with impaired renal function.

The NRTIs require an activation process by the host cell nucleoside kinases (3 phosphorylations for nucleosides and 2 for nucleotides).

The figure (on the left) shows that Zidovudine (ZDV or AZT) is incorporated in the growing chain. The absence of free –OH group at the 3'end (contains an azido group) causes chain termination.



The NRTIs can also inhibit human mitochondrial DNA polymerase gamma (enzyme responsible for mitochondrial DNA synthesis). **Hence, side effects common to the NRTI class is mitochondrial toxicity, often clinically manifested as myopathy.**

As compared to the other NRTIs, cardiomyopathy is seen most often with AZT (Zidovudine). Long term Zidovudine administration also causes bone marrow suppression (e.g. anemia, leukopenia). Other mild complications, observed with all of the NRTIs, include weight loss, fatigue, abdominal pain, and exercise-induced dyspnea. Severe complications may include Septic shock-like syndrome, hepatitis, pancreatitis and lactic acidosis. Due to high mortality rates in patients manifesting these complications, all antiretrovirals are stopped upon induction of these severe side effects.

Both neuropathy and pancreatitis are frequently observed with D4T (stavudine) and DDI (didanosine). Also, patients on combination d4T and ddl show higher incidence of lactic acidosis and hepatic steatosis, hence this combination is not recommended. Women, especially those who are overweight, are particularly at risk of these severe complications of NRTIs.

Abacavir, another NRTI, causes severe hypersensitivity reaction in some patients which is manifested as flu-like illness (fever, myalgias, diarrhea, rash). In patients manifesting these symptoms, **rechallenging with abacavir is contraindicated**, since this can be fatal.

The combination of zidovudine and stavudine is also contraindicated due to antagonism.

NtRTI: **Tenofovir (Viread®)**, the only available nucleotide (nucleoside monophosphate) analog, It is often used in HAART combinations along with another NRTI. Tenofovir's anti-HIV efficacy can be achieved much more rapidly since this NtRTI does not need the initial activation step (rate limiting step) required for the NRTIs. However, due to its negative charge (phosphate group) it does not cross lipid membranes easily (e.g. blood-brain-barrier). Thus, its pharmacokinetic efficacy in HIV reservoirs is significantly lower than the other NRTIs.

Side effects seen with Tenofovir include upset stomach, diarrhea, vomiting, flatulence, and loss of appetite. Tenofovir can cause acute renal failure, Fanconi syndrome, proteinuria, and tubular necrosis. It is cleared via the kidneys, hence tenofovir does not compete with most other anti-HIV drugs that are metabolized by the liver. When used in combination with other anti-HIV drugs, tenofovir has been found to work in patients who have developed resistance to NRTI drugs, especially HIV that is resistant to lamivudine (Epivir, 3TC). However, before adding tenofovir, a genotypic drug resistance test for AZT resistant HIV (with the K65R mutation) is needed. It is the only known mutation that may cause Tenofovir to be less effective. With the exception of didanosine (ddl, Videx), Tenofovir does not affect the level of other drugs in the body. Tenofovir can interact with didanosine by increasing didanosine's concentration.

Decreased pill burden has been achieved with availability of **combination RTI pills**:

Tenofovir is also available in a fixed-dose combination with emtricitabine (FTC) with the brand name Truvada®, for once-a-day dosing. A regimen of atazanavir (Reyataz) + ritonavir (Norvir) + Truvada (tenofovir/FTC) are regularly used. In addition, a new three-in-one pill, Atripla (tenofovir/emtricitabine (FTC)/efavirenz) is also available now-a-days as a complete regimen.

2. Non-nucleoside reverse transcriptase inhibitors (NNRTIs): The mechanism of action of NNRTIs is non-competitive inhibition of the RT enzyme, causing disruption of its function. These drugs do not require an activation process by the cell and have excellent activity against HIV-1, but none towards HIV-2. They have a long half-life which allows for less dosing: Nevirapine: twice/day dosing; Efavirenz: once/day dosing; Delavirdine: three times/day (rarely used; least potent of the class). All three NNRTIs are metabolized through the liver P450 system. It is important to note that both efavirenz and nevirapine are inducers of the P450 system; hence, they may cause decreased levels of others drugs metabolized through this drug metabolizing system.

Toxicities common to the class are rash: nevirapine > delavirdine > efavirenz. From self-limited rash to fatal Stevens-Johnson syndrome (severe muco-cutaneous rash with systemic manifestations) is seen upon long-term use.

Do not use another NNRTI when history of severe rash to any member of the class is seen. Hepatitis is more often seen with Nevirapine, usually observed within first 12 weeks after initiation of therapy, which can be accompanied with rash, flu-like illness.

Efavirenz is **teratogenic** and should be avoided in women of childbearing age. Efavirenz also has CNS side effects which range from light-headedness to hallucinations which decreases with time.

B. Protease inhibitors (PIs):

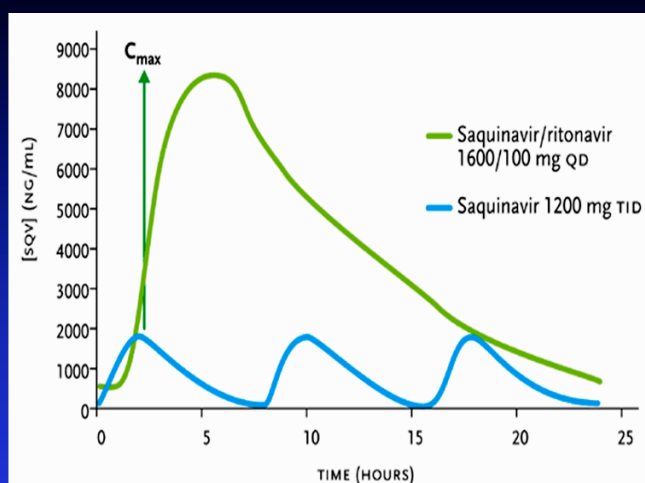
The mechanism of action of PIs is inhibition of the viral protease enzyme (cleaves viral polyproteins, a process needed to produce mature proteins for assembly of the virus). This leads to the assembly of defective, non-infectious, viral particles. The PIs are metabolized through the liver P450 system so drug-interactions is a significant issue.

There are several toxicities which are common to the class. These include, Hepatitis: Ritonavir > others. Can occur at any time, most frequent in patients with other causes of liver disease (hepatitis B or C, alcohol abuse); may be due to immune reconstitution (increased inflammatory response to hepatic injury). Hyperglycemia: Due to insulin resistance/glucose intolerance: seen at 2 weeks in non-HIV infected patients when exposed to PIs. Can lead to new onset diabetes mellitus (DM) (mostly in patients with other risk factors) or to worsening of pre-existing DM. Hypertriglyceridemia/hypercholesterolemia: ritonavir >> others; atazanavir < others. Gastrointestinal effects: dyspepsia, bloating, nausea, vomiting, diarrhea: common to all; usually improves with time: Ritonavir high dose >> others. There are also specific toxicities common to each of the PIs: Indinavir: Renal toxicity, ranges from interstitial nephritis to nephrolithiasis. Amprenavir: Rash (this drug is a sulfonamide, may have cross-reactivity with sulfa drugs).

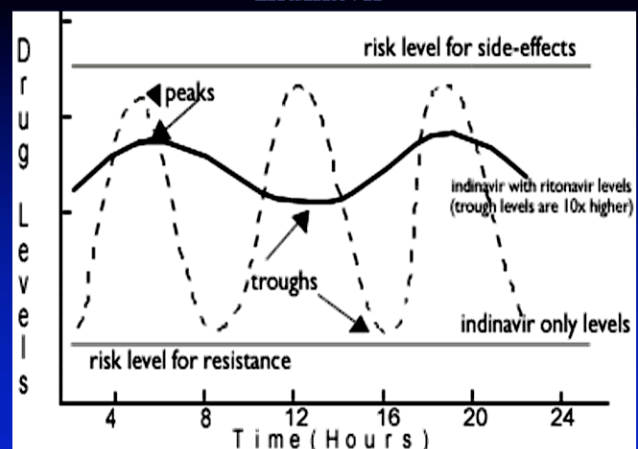
Another important point to note is that all of the PIs are also inhibitors of the P450 system (ritonavir >>> others). Hence, they have important drug interactions with other drugs metabolized through same mechanism (producing increased levels that may be clinically relevant and require dose adjustment). The older members of the class had higher pill burden (saquinavir, indinavir, nelfinavir); however, the newer drugs, e.g. lopinavir, atazanavir and darunavir, have much longer half life.

The boosted PI concept: Currently ritonavir is used not for its antiviral activity (due to its major side effects at full dose) but for its pharmacokinetic effects. Since ritonavir strongly inhibits CYP450, it also inhibits the metabolism of other PIs., its concomitant use at low dose allows for reduction in the dose (less pills and/or less frequency) of other PIs→ritonavir-boosted dose. Thus, the serum levels of PIs achieved with ritonavir-boosted regimens are much higher than those achieved with unboosted regimens. All PIs, except nelfinavir, can be administered in a ritonavir-boosted regimen (preferred regimen for most PIs). **Lopinavir/ritonavir (Kaletra™)** is the only co-formulated capsule; for all other boosted regimens, ritonavir is added as an extra drug (i.e.: saquinavir + ritonavir, atazanavir + ritonavir).

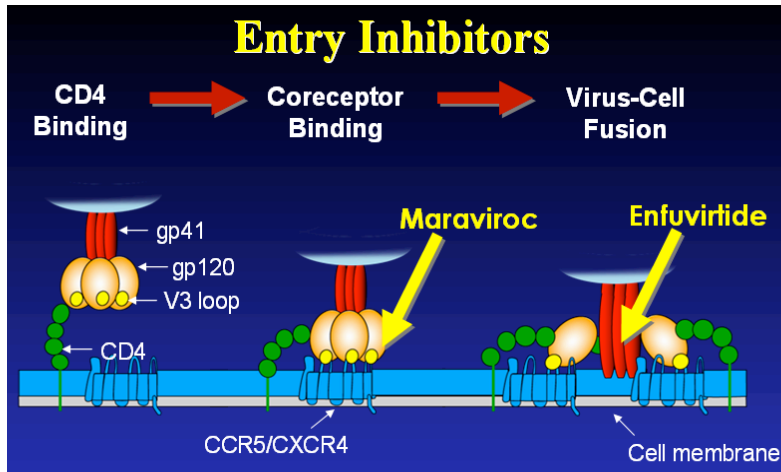
Effect of Ritonavir boosting on peak Saquinavir levels



Effect of Ritonavir Boosting on Serum Levels of Indinavir



C. Entry Inhibitors:



There are currently two classes of entry inhibitors:

The first class interferes with entry of HIV into CD4+ cells by inhibiting the fusion of viral and cellular membranes. Enfuvirtide is the only drug available in this class.

The second class of entry inhibitors are HIV coreceptor antagonists. The clinically approved drug, Maraviroc is a CCR5 antagonist. Currently there are several CXCR4 antagonists in clinical trial.

Enfuvirtide is a very large peptide. It is only available as a subcutaneous (parenteral) formulation, with twice a day (BID) dosing. However, new formulations are in the pipeline. It has potent effect against HIV-1, but since it is very expensive, it is not often used as a component of HAART. It is usually kept as an agent for **salvage therapy** in patients with HIV resistant to both NRTIs and PIs. Enfuvirtide is metabolized through the liver (proteolytic hydrolysis, not through P450 system). Toxicities associated with enfuvirtide include injection site reaction: induration, redness (in 90% of patients) which is usually mild. Interestingly, within a short time after the introduction of this anti-HIV drug into the market, resistance mutants have already emerged.

Maraviroc (Selzentry) is a CCR5-blocking entry inhibitor. It is available in 150 mg and 300 mg tablets and must be taken twice a day and must be combined with other HIV drugs. Maraviroc is prescribed to individuals who have HIV strains that are resistant to multiple antiretroviral drugs. Maraviroc is metabolized by the liver and has no known "contraindications." Anticonvulsants (such as carbamazepine, phenobarbital, and phenytoin), can decrease the amount of maraviroc. Side effects associated with maraviroc include cough, fever, colds, rash, muscle and joint pain, stomach pain, and dizziness. An important point to be noted is that maraviroc will only be effective at reducing viral load in people with HIV that uses the CCR5 receptor.

D. Integrase Inhibitors:

Raltegravir (MK-0518, brand name **Isentress**) targets integrase, an HIV enzyme that integrates the viral genetic material into human chromosomes. It is approved only for use in individuals whose infection has proven resistant to other HAART drugs (salvage therapy). However, raltegravir has been associated with high performance against HIV-1 in treatment-naïve and limited-treatment option patients. It is believed that this enhanced efficacy is potentially because of its binding interaction with the HIV pre-integration complex. In clinical trials, patients taking Raltegravir achieved viral loads less than 50 copies per millilitre sooner than those taking similarly potent NNRTIs or PIs. Research into Raltegravir's ability to affect latent viral reservoirs and possibly aid in the eradication of HIV is currently ongoing.

Raltegravir is taken orally, twice daily, with or without food. Doses of 200, 400, and 600 mg have been used. Most common side effects have been diarrhea, nausea, headache, and fever. Some people have experienced rash, Stevens-Johnson syndrome, depression, and suicidal

tendencies. The drug is metabolized via glucuronidation and not via the CYP450 system. Hence, no dose adjustment of raltegravir is required when it is coadministered with other antiretroviral agents. However, raltegravir should be used with caution when administered with strong inducers of uridine diphosphate glucuronosyltransferase (UGT1A1), including rifampin. These inducers of UGT1A1 may reduce plasma concentrations of raltegravir. Clinical trial data suggested that concomitant use of raltegravir and atazanavir (a strong inhibitor of UGT1A1) boosted with ritonavir caused increased plasma concentrations of raltegravir.

Complications of treatment

In addition to the drug-specific and class-related toxicities previously outlined, there are a number of significant complications seen with the combination HAART regimen. The etiology of these complications is multifactorial, and the following have been implicated. Protease inhibitors may cause an insulin resistance syndrome (IRS) and the NRTIs can cause mitochondrial toxicity. In addition, HIV infection can precipitate immunosuppression. Thus, long-term use of HAART drugs has been linked to a significant increase in the incidence of **lipodystrophy syndrome and cardiovascular disease**.

The lipodystrophy syndrome is characterized by one or more of the following symptoms, such as lipoatrophy (fat wasting): loss of subcutaneous fat, especially proximal upper and lower extremities and face; and hyperadiposity (fat accumulation): Deposition of visceral fat manifested by increased abdominal girth, and/or abnormal deposition of fat elsewhere (buffalo hump, lipomatosis, anterior neck). Also, in both men and women, Gynecomastia: Increased breast size due to deposition of fat, is often seen.

Increased risk of cardiovascular disease is apparent in several cohort and observational studies in long-term HAART users. This has been attributed to the increased incidence of atherogenic lipid profile (high LDL, low HDL, increased triglycerides) and the PIs appear to be the main culprits. Main intervention is to avoid the PI containing HAART regimen and treat other risk factors, such as smoking cessation, exercise, and low-fat diet. In addition, treatment of hypertension and hyperlipidemia using ACE-inhibitors and Statins, are also recommended in these patients.

All classes of antiretrovirals have been associated with hepatitis, especially among patients with underlying liver disease (Hepatitis C). Both NNRTIs and PIs affect the metabolism of other drugs metabolized through the P-450 system and **have been associated with significant drug interactions**. Hence, always check when adding a new drug to a patient on antiretrovirals or when starting antiretrovirals on a patient on other drugs.

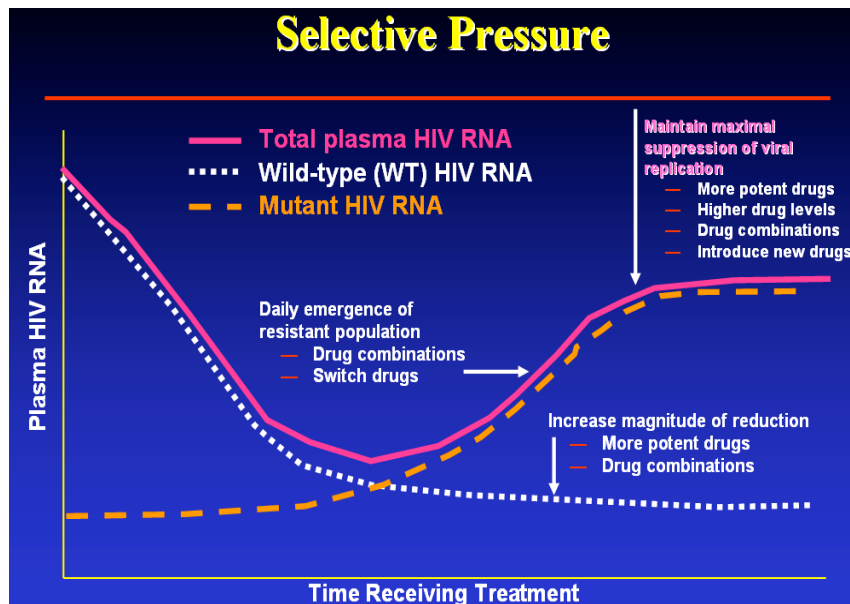
Viral reservoirs and HAART

With untreated infection, most patients have high levels of viral replication. Combination therapy that includes a PI or NNRTI and 2 NRTIs causes potent inhibition of viral replication. With potent combination therapy (HAART), a quick drop in viral load is seen at 2-4 weeks (>1 log), and maximal response is achieved at 12-24 weeks. With maximal viral suppression, the viral load drops below the level of detection (<400, <50 or as low as <5 RNA copies/mL depending on the test used). In patients with initially susceptible virus and high-level adherence, suppression is long lasting. However, even in patients with long-lasting, maximal suppression for >5 years the virus is not eradicated (it persists in resting lymphocytes and other long-lived infected cells), and low-level viral replication occurs. With currently available antiretrovirals, HIV replication can be maximally suppressed, but **the virus cannot be eradicated**.

HAART has turned HIV infection into a chronic, treatable condition, similar to other chronic diseases such as hypertension and diabetes. However, the main reason for lack of long-lasting viral suppression is the **emergence of resistant virus**. Latent HIV-1 reservoirs, in memory T-cells as well as those infected cells sequestered within tissues, e.g. bone marrow, lymph nodes and the CNS, can play a significant role in viral replenishment and selection of drug resistance.

Viral drug resistance

With untreated infection, most patients have high levels of viral replication. The viral reverse transcription process is highly prone to error, causing an average of 1 mutation/viral cycle. As a consequence, in any one patient there is a high level of viral heterogeneity and a diverse mixture of HIV-1 quasiespecies. Consistent levels of drug above the IC50 (inhibitory concentration of 50% of



the virus in a culture system) are essential for viral suppression. In the face of sub-inhibitory concentrations, resistant virus can emerge. In the absence of maximal suppression of viral replication, the chances of emergence of mutations that would confer resistance to the medication(s) to which the virus is exposed are high. Interestingly, when drug pressure stops (patient is off medications) viral population tends to shift from resistant virus to wild type (susceptible) virus. But **resistant virus is not gone, it is archived, and it will emerge with re-exposure to the drug**.

Regimens in which drug levels are well above viral inhibition levels throughout the dosing interval are more forgiving of missed doses. Thus, the advantage of ritonavir-boosted regimens.

For some drugs (lamivudine and NNRTIs), one viral mutation is sufficient to cause high-level resistance to those drugs. Other drugs require accumulation of several mutations to produce high-level resistance. **Cross resistance among members of a drug class is observed often**. With NRTIs, different mutations produce cross-resistance among different drugs, and multi-drug resistant mutations have been identified. With NNRTIs, the relevant mutation produces cross-resistance among all members of the class. With the PIs, different mutations are relevant for different PIs, and accumulation of mutations yield cross-resistance. Hence, combinations of drugs are indicated to avoid emergence of resistance (suppressed virus cannot generate all the mutations needed to produce resistance to all drugs).

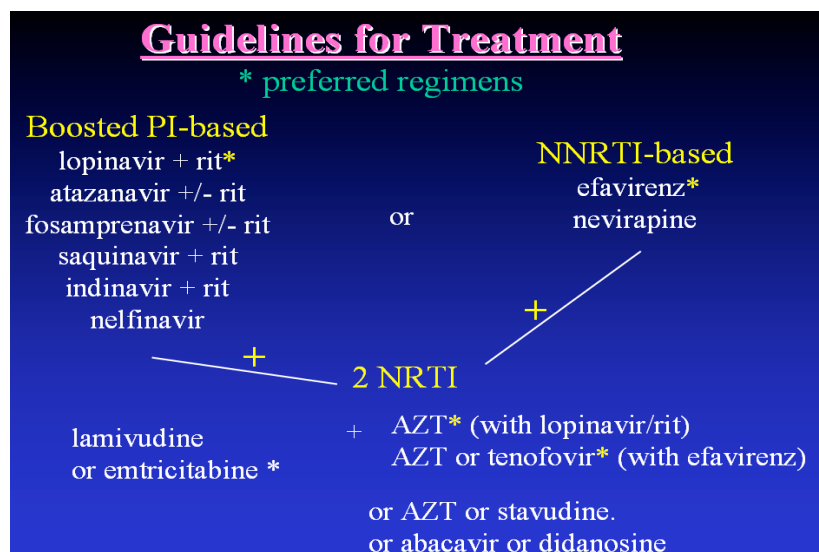
Testing for resistance:

- **Resistance testing (whenever possible) should be obtained while the patient is on treatment.**

- Phenotyping assays: measures the patient's virus ability to grow in the presence of different concentrations of the drug. Growth in the presence of the relevant concentration of drug indicates resistance.
- Genotyping assays: The mutations associated with resistant virus have been identified by sequencing. The presence of these mutations in the patient's virus indicates resistance.

Adherence to treatment is the most important factor to prevent resistance. Lack of adherence is the most important reason for serum sub-inhibitory levels of drug and the most important determinant for emergence of resistance.

Guidelines for initiation of treatment



Guidelines are issued by different agencies in different countries. In the USA, clinicians follow the guidelines of the U.S. Public Health Service and Department of Health and Human Service (DHHS). Guidelines and regular updates are available at: <http://AIDSinfo.nih.gov>.

Treatment recommendations change as more data becomes available regarding efficacy and toxicities, and as new drugs that are easier to take and/or are associated with less toxicity become available.

Guidelines are intended to pursue the following **Goals of treatment**:

- Maximal and durable suppression of viral load
- Restoration or preservation of immunologic function
- Improvement in quality of life
- Reduction of HIV-related morbidity and mortality

The decision on **when to start treatment** is based on evaluation of factors known to be important in determining risk of progression of disease (AIDS). Clinical status of the patient is a key issue and all symptomatic patients need treatment (there is no controversy regarding this issue). The Viral load is another marker of treatment initiation. Higher viral loads mean more risk for progression to AIDS. The CD4 cell count of the patient is another marker for risk of progression. Outcome is the same when treatment is started at CD4 cell counts of >500 or at 350-500. However, outcome is not as good when treatment is started at <250. The CD4 count range of 200-350 continues to be gray area when discussing indications for treatment.

Management of HIV-infected patients

All patients on treatment should have the following laboratory evaluations:

- Viral load: 4-6 weeks after initiation of treatment (to assess initial response), at 3 months (to assess maximal suppression), and every 3-4 months (to assess durability of response).
- CD4 cell count should be done to assess immune reconstitution post treatment.

Treatment failure (lack of viral suppression) occurs when viral load of <400 not achieved after 3-6 months of HAART. Increased viral load after initial suppression indicates **virologic failure**. This is usually secondary to selection of resistant virus.

Changing antiretrovirals:

When starting new drugs on a patient that has failed an initial antiretroviral regimen, effort should be made to include at least 2 new drugs (and if possible 3) to which the virus is susceptible.

Salvage therapy: Antiretroviral treatment for patients who have failed multiple regimens and who carry virus that is resistant to all or most classes of drugs (multi-drug resistant virus or MDR). Entry inhibitors and integrase inhibitors have improved the response to salvage therapy. Some patients will achieve complete viral suppression with salvage therapy that includes one of these newer anti-HIV agents.

Mega-HAART: This is a regimen of over 5 drugs (2 PIs, 2 NNRTIs, 3 or 4 NRTIs, and hydroxyurea used together have been reported). However, toxicity and drug-drug interactions are limiting factors.

Special Treatment Considerations:

Primary HIV-1 infection: When patients are diagnosed with acute retroviral syndrome or within 6 months of HIV infection, consideration can be given to initiate therapy.

Pregnancy: Pregnant women should be treated following current guidelines.

- If initiating therapy, consider delaying antiretrovirals during the first 10-12 weeks of pregnancy.
- **Avoid efavirenz (teratogenic) throughout pregnancy.**
- Use approved protocols for prevention of transmission to newborn.

Children: A different document "Guidelines for the use of antiretroviral agents in pediatric HIV infection" is issued by DHHS. Specific issues in children:

- Acquisition of infection through perinatal exposure for many infected children
- Age-specific differences in immunologic parameters (i.e., CD4 cell counts)

Adolescents: Medications dosages should be based on Tanner staging of puberty and not specific age.

- Pediatric doses for those in early puberty.
- Adult doses for those in late puberty.